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Age-Dependent Accuracy of *Helicobacter pylori* Antibody Assays for Adults, with Special Emphasis on Atrophic Gastritis

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The accuracy of *Helicobacter pylori* antibody assays for 561 consecutive adult outpatients who had undergone gastroscopy was studied. The sensitivity of an immunoglobulin G test was 99 to 100% for all age groups, but the specificity declined by age group, from 99% for those aged 15 to 49 years to 75% for those aged ≥65 years. The exclusion of false-positive results for patients with atrophic gastritis improved the specificity to 93 to 97% for the older age groups.

Helicobacter pylori infection is associated with gastritis, peptic ulcer disease, and gastric malignancies (1). In about one-third or up to one-half of those infected with *H. pylori*, gastritis proceeds to atrophic gastritis, resulting in a loss of mucosal glands, decreased helicobacter colonization and, when affecting the corpus mucosa, decreased secretion of pepsinogen I (PGI) (10, 23).

Most helicobacter-infected subjects have specific circulating immunoglobulin G (IgG) antibodies. However, IgA antibodies are found in approximately two-thirds of infected subjects (9). Of infected individuals, 2 to 7% show an elevated level of IgA antibody only (5, 9). Patients with atrophic corpus gastritis often have positive helicobacter serology, although microscopic examination (6, 22), culture of biopsy samples, and even the urea breath test remain helicobacter negative (8). These particular patients may still be infected, as shown by rapidly falling antibody titers after therapy (7).

Enzyme immunoassay (EIA) is the most commonly used serological method for detecting antibodies to *H. pylori*. The best commercial kits have shown sensitivities and specificities of 90% to in excess of 95% (3, 12). The present study focuses on the diagnostic accuracy of *H. pylori* IgG and IgA antibody tests for adults in different age groups, with special emphasis on the presence of atrophic gastritis.

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Patients. We analyzed the data from 561 consecutive adult outpatients (age range, 16 to 91 years; median age, 56 years; 60.4% [339] female), who had undergone gastroscopy due to clinical indications at the primary care level at Vammala Health Center, Vammala, Finland, from December 1998 to November 2002. Patients who had been treated successfully for helicobacter infection were excluded.

Biopsy samples. Gastroscopies were performed by one of the authors (A.S.-R.) in a routine manner, with two biopsies from the gastric antrum (2 cm or more from the pylorus), two biopsies from the large curve of the gastric corpus for histological examination, and additional biopsies (one from the antrum and one from the corpus) for culture.

Formalin-fixed biopsy specimens were embedded in paraffin. Tissue sections of 3 μm were cut at three levels per biopsy and placed on one slide. The tissue sections were stained with hematoxylin and eosin, Alcian blue (pH 2.5)–periodic acid-Schiff, and modified Giemsa. Gastritis was classified according to the Sydney System, in a blinded fashion, by one pathologist (J.M.) (20).

The biopsies were mailed for culture in Transpocult tubes (Orion Diagnostica, Espoo, Finland). The specimens were cultured for *H. pylori* for up to 12 days on Brucella agar plates (Becton Dickinson, Sparks, Md.) (18) supplemented with whole horse blood (7%) and on selective Brucella agar plates containing Iso-Vitalex (1%), vancomycin (6 mg/liter), amphotericin B (2 mg/liter), and nalidixic acid (20 mg/liter).

Results were considered helicobacter positive if culture or histology or both were positive.

Serum tests. Serum samples were taken during each patient's visit to the endoscopic unit and were stored at −20°C until analyzed. Sera were tested for *H. pylori* IgG and IgA antibodies and PGI levels (16) by using commercially available EIAs (Pyloriset EIA-G III, Pyloriset EIA-A III, and Gastroset PGI; Orion Diagnostica). According to the manufacturer's instructions, PGI values below 28 μg/liter were considered low. After validation (results not shown) of Pyloriset EIA-G III and Pyloriset EIA-A III tests, titers of 30 or higher were considered positive instead of the cutoff value of 20 recommended by the manufacturer.

Statistics. The sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were determined for each test and compared with the prevalence of *H. pylori* based on histology or culture or both. The statistical significance of the improvement of the specificity figures was

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TABLE 1. Endoscopic findings for the 561 consecutive patients referred for upper endoscopy

Finding	No. of patients with indicated *H. pylori* test result	
	Positive (n = 181)[a]	Negative (n = 380)[b]
Normal gastric mucosa	0	206 (54.2)
Hiatus hernia only	27 (14.9)	87 (22.9)
Erosive esophagitis[c]	43 (23.8)	69 (18.2)
Barrett's esophagus	3 (1.7)	8 (2.1)
Erosive gastritis	5 (2.8)	18 (4.7)
Peptic ulcer disease[d]	22 (12.1)	5 (1.2)[e]
Gastric malignancy	1 (0.5)[f]	4 (1.0)[g]

[a] Culture or histology or both were positive.
[b] Culture and histology were both negative.
[c] Includes 10 *H. pylori*-positive and 27 *H. pylori*-negative patients with hiatal hernia.
[d] Includes 8 patients with duodenal ulcers and 3 patients with gastric ulcers among *H. pylori*-infected patients, 12 patients with ulcer scars (9 *H. pylori*-positive and 3 *H. pylori*-negative patients), and 4 patients with previous antrectomies due to peptic ulcer disease (2 *H. pylori*-positive and 2 *H. pylori*-negative patients).
[e] $P < 0.0001$.
[f] Includes one patient with adenocarcinoma.
[g] Includes three patients with adenocarcinomas and one patient with lymphoma.

examined by using Pearson chi-square and Fisher's exact tests. The trend in proportion by age was evaluated by using a linear-by-linear test. Statistical analyses were done by using StatView 5.0 and SPSS 12.0 software packages (SPSS, Inc., Chicago, Ill.).

Presence of *H. pylori* and atrophic gastritis. Endoscopic findings are shown in Table 1. Of the 181 *H. pylori*-positive patients, culture was positive for 173 (95.6%), while microscopy of the histological preparations revealed the presence of the bacterium for 161 (88.9%) ($P = 0.0305$). Of the patients with a positive culture result without helicobacter in histological examination, all 19 demonstrated chronic gastritis and 12 showed atrophic changes in the gastric mucosa.

Of the 181 patients with *H. pylori* gastritis, 66 (36.5%) showed atrophic gastritis in histology, while 54 (14.2%) of the 380 *H. pylori*-negative patients demonstrated atrophic gastritis in biopsy specimens ($P < 0.0001$). The grades of atrophic changes in the corpus mucosa and their relationship to low PGI values are shown in Table 2. Of the 24 *H. pylori*-infected patients with atrophic changes in the antrum mucosa, 17 (70.8%) demonstrated low-grade atrophic changes and only 2 patients showed advanced severe atrophy in the antrum mucosa. Similarly, 7 (70%) of the 10 *H. pylori*-negative patients with atrophic changes in the antrum mucosa had low-grade changes and no patients had severe changes. Atrophic gastritis based on histology or a low PGI value was more common in the older age groups, appearing in 10.2, 17.2, and 39.6% of the patients aged 15 to 49, 50 to 64, and >65 years, respectively ($P = 0.0001$).

***H. pylori* antibodies.** In young adults with a low prevalence of *H. pylori* infection, the IgG test demonstrated very high sensitivity and specificity figures (100 and 99%, respectively) and PPVs and NPVs (95 and 100%, respectively) (Table 3). In the older age groups, the sensitivity of the IgG test was high (over 98%). The number of false-positive results increased with patient age, resulting in a decrease in specificity to 75% for patients aged 65 years or older. After exclusion of the patients with false-positive antibody values and atrophic gastritis, the specificity for the older age groups increased to 93 to 97% (Table 3).

The sensitivity of the IgA test increased and the specificity of the IgA test decreased with patient age (Table 3). For young adults the specificity was 97%. After exclusion of the false-positive results for patients with atrophic gastritis, the specificity was 92 to 95% for older age groups (Table 3).

Comments. In this study, we applied a novel approach to observe the effect of increasing age and presence of atrophic gastritis on the diagnostic performance of *H. pylori* IgG and IgA tests. The IgG test was highly sensitive in adult patients, which makes it an excellent choice for the test-and-treat approach recommended for young adult dyspeptic patients (15, 21). Both tests demonstrated high specificity in subjects below

TABLE 2. Elevated *H. pylori* antibody levels of the IgG class, atrophic corpus gastritis, and low serum pepsinogen I in *H. pylori*-positive and -negative patients

Test result	No. of patients with the indicated grade of corpus atrophy[a]				Total no. of patients
	0	1	2	3	
H. pylori positive					
IgG positive[b]	129	31	14	3	177
Histology positive[c]	123	28	6	3	160
Culture positive[d]	126	30	12	4	172
Histology and/or culture positive	130	31	14	4	179[e]
Low PGI (%)[f]	1 (0.8)	1 (3.2)	4 (28.6)	1 (25)	7 (3.9)
H. pylori negative					
IgG positive[b]	18	6	8	7	39
Histology and culture negative	328	13	16	22	379[e]
Low PGI (%)[f]	10 (3.0)	4 (30.8)	10 (62.5)	21 (95.4)	45 (11.9)

[a] Corpus atrophy was graded according to the Sydney System.
[b] Elevated *H. pylori* IgG levels (≥30 U/ml) by Pyloriset EIA-G III.
[c] *H. pylori* infection was present based on histology of antrum and corpus mucosa.
[d] *H. pylori* infection was present based on culture of antrum and corpus mucosa.
[e] There were two *H. pylori*-positive patients and one *H. pylori*-negative patient with superficial biopsy specimens without glands, which explains the three missing patients in this total.
[f] Low PGI levels (<28 μg/liter) by Gastroset PGI EIA.

TABLE 3. Sensitivities, specificities, PPVs, and NPVs of *H. pylori* EIA antibody tests for different age groups for all patients and after excluding patients with atrophic gastritis[a]

Age group (yr) and prevalence (%)[b]	No. of patients	Test	Sensitivity	Specificity	PPV	NPV	*P* value for increase in specificity
15–49	157						
H. pylori+ 13.4		IgG	100 (100)	99.3 (99.3)	95.4 (95.4)	100 (100)	
Histology+ 12.1		IgA	52.4 (52.4)	97.1 (97.1)	73.3 (73.3)	92.9 (92.9)	
Culture+ 13.4		IgG + IgA[c]	100 (100)	96.3 (96.3)	80.8 (80.8)	100 (100)	
50–64	192						
H. pylori+ 37.5		IgG	98.6 (98.6)	94.2 (97.4)	91.0 (95.9)	99.1 (99.1)	NS
Histology+ 33.3		IgA	80.6 (80.6)	91.7 (94.8)	85.3 (90.6)	88.7 (88.7)	NS
Culture+ 36.5		IgG + IgA	98.6 (98.6)	89.2 (93.0)	84.5 (89.9)	99.1 (99.1)	NS
≥65	212						
H. pylori+ 41.5		IgG	98.9 (98.9)	75.0 (93.0)	73.7 (92.5)	98.9 (98.9)	<0.0001
Histology+ 36.8		IgA	86.4 (86.4)	79.0 (92.4)	74.5 (90.5)	89.1 (89.1)	0.0043
Culture+ 38.7		IgG + IgA	100 (100)	70.2 (86.1)	70.4 (86.3)	100 (100)	0.0044
All	561						
H. pylori+ 32.3		IgG	98.9 (98.9)	89.7 (96.9)	82.1 (82.1)	99.4 (99.4)	<0.0001
Histology+ 28.7		IgA	80.1 (80.1)	89.5 (95.0)	78.4 (88.9)	90.4 (90.4)	0.0055
Culture+ 30.8		IgG + IgA	99.4 (99.4)	85.5 (NC)	76.6 (NC)	99.7 (NC)	NC

[a] Results after exclusion of patients with atrophic gastritis and a false-positive antibody result are shown in parentheses. Atrophic gastritis was verified by histology or a Pepsinogen I value of <28 μg/liter or both. NS, nonsignificant. NC, not calculable because of different numbers of false-positive subjects in IgG and IgA tests.
[b] *H. pylori*+, prevalence of *H. pylori* was based on histology or culture or both; histology+, prevalence of *H. pylori* was based on histology alone; culture+, prevalence of *H. pylori* was based on culture alone.
[c] Combination of IgG and IgA results.

50 years of age, with a low prevalence rate of *H. pylori* infection but a lower performance in older subjects due largely to atrophic gastritis. However, the specificity improved clearly after older patients with atrophic gastritis and false-positive antibody results were excluded. In addition, in serologic assays, the prior use of antibiotics, antisecretory treatments, or the location or the reduced number of helicobacters on the gastric mucosa had no effect on the diagnostic accuracy (4). However, antisecretory treatments before gastroscopy may lead to false-negative results for histology, culture, and urease tests (2, 14). It is also known that *H. pylori* is absent from areas of intestinal metaplasia, a common finding with chronic gastritis (19).

The sensitivities and specificities of the commercial *H. pylori* antibody tests seemed to vary between 60 and 100% (3, 12). The accuracy of serological tests is strongly dependent on the prevalence of *H. pylori* infection (13, 17). Although it has been recommended that antibody assays be evaluated locally (4, 13), this has rarely been carried out for different age groups. Our results of the accuracy of the *H. pylori* IgG and IgA tests were superior to those reported earlier (3, 11, 12, 24). To avoid any misjudgment in the validation of serological tests for *H. pylori* antibodies in adult subjects, the validation should be carried out separately for different age groups with special emphasis not only on the known *H. pylori* status but also on the presence of atrophic gastritis.

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